Spontaneous calcium (Ca) waves must emerge near-synchronously in thousands of contiguous myocytes to produce delayed afterdepolarizations (DADs) in cardiac tissue. Previous studies have shown as Ca load increases, the time to onset of a spontaneous Ca wave after pacing (latency), as well as its variability, decreases. Two proposed mechanisms include the time period required to refill the sarcoplasmic reticulum (SR) Ca stores and regain Ca release channel excitability, and the latter plus an ''idle period.'' Here we used patch-clamped Fluo-4-loaded isolated rabbit ventricular myocytes to detect Ca waves and DADs following pacing trains. Longer pacing trains enhancing Ca loading decreased latency and its variability. Using paced premature beats, latency outlasted the period required for SR refilling and Ca release channel recovery, consistent with an additional ''idle period.'' From combined experimental data, simulations, and theoretical analysis, we present evidence that the ''idle period'' can be explained by criticality theory (Biophys J 2012;11:2433), related to the probability that a cluster of Ca release units large enough to initiate a Ca wave will self-organize. This theory directly accounts for the observed shortening of latency and its variability as Ca load increases.

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Contributions of I(F) and Sarcoplasmic Reticulum Ca^{2+} in the Control of Spontaneous Cardiac Beating Rate in Mouse and Guinea-Pig

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Spontaneous beating rate was studied in mouse isolated atrial preparations and in guinea-pig single myocytes isolated from the sinus node (SAN) region of the heart. In atrial preparations, the I(f) blockers ZD7288 (1 μM) and ivabradine (3 μM) both reduced spontaneous beating rate by more than 200 bpm, and the change in beating rate caused by the combination of ivabradine and ZD7288 was not significantly different from the changes caused by either drug alone. Application of cyclopiazonic acid (CPA, to inhibit SERCA) also reduced beating rate by about 150 bpm. ZD7288 alone, ivabradine and the combination of ZD7288 and ivabradine all depressed the log(concentration)-effect curves for the effect of isoproterenol on rate by similar amounts. CPA alone caused a greater depression of isoproterenol log(concentration)-effect curves than was the case with the I(f) blockers. The combination of I(f) blockers and CPA caused a profound further depression of the effects of isoproterenol on spontaneous rate so that the log(concentration)-effect curve was almost flat. The effects I(f) blockers and CPA were also investigated in guinea-pig isolated SAN myocytes loaded with the Ca^{2+} probe, fluo-5F, and were broadly comparable to those observed in intact spontaneously beating atria. The observations are consistent with roles for both I(f) and SR Ca^{2+} in the control of spontaneous beating rate. The balance between the contributions of different mechanisms to the control of spontaneous rate varies with conditions, particularly in their influence on the increase in rate caused by isoproterenol. The presence and function of Ca^{2+}-stimulated adenylyl cyclases is likely to be an important factor with the potential to link the contributions of these interacting processes.

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Doxorubicin Stimulates the Na^+/Ca^{2+} Exchanger in Ventricular Cardiomyocytes

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Doxorubicin (DOX) is a widely used antineoplastic agent that exerts cardiotoxic effects. This study analyzed the effects of a clinically relevant DOX concentration (30 μM) on Ca^{2+} transport in rat ventricular myocytes. DOX decreased the sarcoplasmic reticulum (SR) Ca^{2+} content ($[Ca^{2+}]$SR) (15%, $p<0.001$), however without significant change in Ca^{2+} transient amplitude and kinetics, or in the fraction of $[Ca^{2+}]$SR released at a twitch. DOX abbreviated $[Ca^{2+}]$i decline of caffeine-evoked transients ($p<0.001$). Estimates of the integrated Ca^{2+} fluxes attributable to individual transporters during twitch $[Ca^{2+}]$i decline indicated a small depression of SR Ca^{2+} uptake (12%, $p<0.05$), but great enhancement of Ca^{2+} efflux via the Na^+/Ca^{2+} exchanger (NCX) (90%, $p<0.05$), so that the contribution of the latter to cytosolic Ca^{2+} removal more than doubled. To promote NCX-mediated Ca^{2+} influx in the absence of electrical stimulation, extracellular $[Na^+]$ was lowered from 140 to 105 mM. In these conditions, DOX exposure increased both the rate constant of $[Ca^{2+}]$i rise (12.8 ± 1.4 vs. 4.8 ± 0.8 nM.s-1) and the rate of spontaneous Ca^{2+} transients (0.18 ± 0.04 vs. 0.05 ± 0.01 Hz, $p<0.05$). After SR function inhibition with thapsigargin, spontaneous Ca^{2+} transients were absent, but NCX-mediated increase in $[Ca^{2+}]$i remained greater in the presence of DOX (190 ± 35 vs. 74 ± 18 nM.min-1, $p<0.01$), which was abolished by inhibition of reverse NCX with 10 μM KB-R7943 (-29 ± 6 nM.min-1, $p<0.01$). The results indicate that DOX altered mainly diastolic Ca^{2+} handling, causing mild inhibition of SR Ca^{2+} uptake, but marked increase of Ca^{2+} transport via NCX. The latter effect does not seem to be due to thermodynamic factors, but probably to NCX stimulation, as both Ca^{2+} influx and efflux were enhanced.

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Local Calcium Dynamics Stabilize NCX Current in an Integrated Calcium Cycling and Membrane Model

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It has been suggested that Na-Ca exchanger (NCX) plays an important role in pacemaker function as a link between intracellular Ca^{2+} dynamics and the membrane ion currents. This role remains unresolved due to a lack of specific NCX blockers and the challenges in measuring NCX current (INCX) under different conditions. We approach the problem via a new model that combines a model of SA node cells featuring local Ca^{2+} control with the entire ensemble of ion currents described by an earlier Maltsev-Lakatta common pool model in order to simulate pacemaker potentials. This approach discovered a new mechanism of stabilization of the INCX using local calcium dynamics when the number of NCX molecules is decreased (as for example in an incomplete NCX knock-out). In this model, during the diastolic depolarization, the Ca^{2+} released via a ryanodine receptor (RyR) in the sarcoplasmic reticulum in SANC can recruit its neighboring RyRs to release more Ca^{2+}. The extent of this local RyR recruitment depends upon the extent to which the released Ca^{2+} diffuses and interacts with neighboring RyRs. As the NCX "steals" Ca^{2+} in the vicinity of each RyR, it restrains C2+a induced Ca^{2+} release. This restraint wanes as NCX expression becomes reduced, ensuring that INCX remains almost unchanged. We furthermore examine when this stabilization mechanism becomes saturated, and find that saturation occurs when all RyRs open to release Ca^{2+}. This leads to arrhythmias and pacemaker failure. Comparing our results to earlier common-pool models we find that only ours predicts physiological Ca^{2+} levels as NCX expression is reduced. Thus our simulations discovered a new INCX stabilization mechanism in cardiac pacemaker cells via local Ca^{2+} control.

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Depolarization of Cardiac Membrane Potential Promotes Calcium Waves

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A calcium (Ca) wave propagates when Ca sparks recruit new sparks in the adjacent Ca release units (CRUs) in cardiac myocytes. When Ca releases occur, Ca from the sarcoplasmic reticulum (SR) is removed mostly via the SERCA pump and the sodium-calcium exchanger (NCX) from the cytosol. If this removal process is more dominant than the positive feedback process of Ca induced Ca release, Ca waves cannot propagate. The diastolic membrane potential can be hyperpolarized or depolarized by various factors such as hyperkalemia, hypokalemia in the long term or by delayed afterdepolarizations (DADs) in the short term. In this study we investigate how membrane potential affects Ca waves. We use a physiologically detailed ventricular myocyte mathematical model to investigate individual factors which affect Ca wave propagation. We investigate the voltage range of $-90 \sim -70$ mV. We find that depolarization of the membrane potential promotes Ca wave propagation and hyperpolarization prevents Ca wave propagation. Ca transport by NCX is determined by membrane potential as well as sodium and Ca concentrations. Depolarized membrane potential reduces NCX-mediated efflux, and thus promotes Ca wave propagation. Moreover, depolarized membrane potential promotes spontaneous Ca releases which can cause initiation of multiple Ca waves. This indicates that during DADs, CRUs interact with not just the immediately adjacent CRUs via Ca diffusions, but also farther CRUs via fast (~0.1ms) voltage diffusion on the membrane through the NCX. This may also be an additional mechanism of synchronization of Ca waves among multiple cells in tissue.

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Observing the Dynamics of Luminal and Cytosolic Calcium During IP3R-Mediated Calcium Signals

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Ca^{2+} signaling is ubiquitous across cell types. Ca^{2+} liberation through inositol 1,4,5-trisphosphate receptors (IP3Rs) is a key component of the Ca^{2+} signaling toolkit. The role of cytosolic calcium on the kinetics of IP3R's and on the dynamics of the evoked signals has been studied at large both experimentally